Associations between dual-phase CT features and histopathologic diagnoses in 52 dogs with hepatic or splenic masses

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Abstract

Ability to non-invasively differentiate malignant from non-malignant abdominal masses would aid clinical decision-making. The aim of this retrospective, cross-sectional study was to identify features in dual-phase computed tomographic (CT) studies that could be used to distinguish malignant from non-malignant hepatic and splenic masses in dogs. Medical records were searched for dogs that had an abdominal dual-phase CT examination, a hepatic or splenic mass, and subsequent histopathologic diagnosis. CT images were acquired prior to and <30s (early phase) and >60s (delayed phase) after intravenous contrast administration. Fifty-two dogs with 55 masses were studied: 24 hepatic, including 14 (58%) malignant and 10 (42%) non-malignant; 31 splenic, including 18 (58%) malignant and 13 (42%) non-malignant. There was substantial overlap in the pre- and post-contrast CT features of malignant and non-malignant hepatic and splenic masses. Regardless of histologic diagnosis, hepatic masses most frequently showed marked, generalized enhancement in early phase images that persisted in the delayed phase. Splenic hemangiosarcoma and nodular hyperplastic lesions most frequently showed marked, generalized enhancement in early phase images that persisted in delayed images whereas most splenic hematomas had slight enhancement in early phase images. All splenic hematomas and 77% of the hemangiosarcomas had contrast accumulation compatible with active hemorrhage. There were no other significant differences in quantitative or categorical CT data between malignant and non-malignant hepatic or splenic masses. Dual-phase CT of dogs with hepatic or splenic masses provides limited specific diagnostic information.

Introduction

Hepatic and splenic masses are encountered frequently in dogs and reflect a range of pathological conditions including malignant neoplasia such as hepatocellular carcinoma and hemangiosarcoma, benign neoplasia such as hepatocellular adenoma and non-neoplastic conditions such as hepatic nodular hyperplasia and hematoma.[1-6] Optimal management depends on accurate diagnosis and staging.[7] Ability to non-invasively differentiate malignant from non-malignant masses would aid clinical decision-making.

In humans with a suspected hepatic mass, multi-phase computed tomographic (CT) protocols have been used to examine the pattern of enhancement of hepatic lesions following administration of intravenous contrast medium.[8,9] The purpose of multi-phase CT protocols is to reveal differences in the vascular supply to lesions that reflect features of diagnostic significance. For example, hypervascular hepatic lesions (such as neoplasms with marked neovascularization) derive their blood supply primarily from the hepatic artery, so show relatively marked contrast uptake in early post-contrast CT images.[8-10] Hypovascular lesions (such as metastases with necrosis) generally show reduced contrast uptake compared to the surrounding liver in late post-contrast CT images.[8,9] However, differentiation of malignant and non-malignant hepatic lesions using contrast uptake characteristics is complicated because non-malignant lesions (such as hepatic nodular hyperplasia) may also show signs of early marked contrast uptake followed by reduced uptake in later images, which mimics the appearance of malignant neoplasms.[8]

In conscious humans, peak opacification of the hepatic arteries occurs at 15-25s, peak opacification of the portal veins occurs at 45-55s, and peak opacification of the hepatic veins occurs at 60-70s after the start of contrast medium injection.[8,11] Triple-phase CT protocols, involving images acquired during the early and late hepatic arterial phase and the portal phase

were studied because of the potential for increased accuracy of diagnosis[9,12,13], but it was found that acquiring two arterial phases carried no significant advantage.[14,15] Triple-phase CT protocols are now used less frequently in human clinical practice than a dual-phase approach involving an hepatic artery-dominant phase (HAP) and a portal venous-dominant phase (PVP) with CT images acquired using time delays of 30-35s and 60-65s after the start of intravenous injection, respectively.[8,14-17] In dual-phase CT protocols to examine the liver, images are acquired after peak vessel opacification because contrast accumulation in parenchymal lesions occurs after peak vessel opacification.[8,11,14] Additional delayed phase images at 180s are recommended by some authors[18] but are considered low-yield by others.[19] In general, HAP images aid detection of hypervascular hepatic lesions[8,12-16] whereas PVP images and delayed images help characterize certain hepatic lesions, such as hemangiomas, and aid detection of active hemorrhage.[20]

Dogs with hepatic masses have not been investigated using dual-phase CT. Instead, triple-phase CT studies have been described, with images acquired at 13-20s (HAP), 30-40s (early PVP), and 120s (equilibrium or delayed phase) following contrast administration.[21,22] These studies found evidence of associations between the pattern and degree of post-contrast enhancement and the pathologic diagnosis of hepatic masses, although there was substantial overlap.[21,22] Most hepatocellular carcinomas showed heterogeneous enhancement in HAP, PVP and delayed phase images, often had a central zone of hypoattenuation, and were hypoattenuating compared to the liver in portal and delayed phase images.[21,22] Hepatic adenomas were more likely to show diffuse enhancement in HAP images and to retain contrast in PVP images.[21] Hepatic hyperplastic nodules were relatively homogeneous, frequently showed marked enhancement in HAP and PVP images, and isoattenuation compared to adjacent normal liver in delayed phase images.[21,22] In

one study, hepatic metastases consistently appeared homogenous with hypoattenuation compared to the liver in all post-contrast CT images.[22]

Multi-phase CT imaging of canine splenic masses has not been reported. A single-phase CT study of splenic masses in dogs used images acquired prior to and approximately 60s after intravenous contrast administration.[23] It was found that malignant lesions, mainly hemangiosarcoma, had lower attenuation than non-malignant masses in pre-contrast images and enhanced less than surrounding splenic parenchyma in post-contrast images.[23]

The aim of the present study was to review post-contrast CT images acquired using a dual-phase protocol in a series of dogs with histopathologically-confirmed hepatic or splenic masses in order to identify features that could be used to distinguish malignant from non-malignant masses. The term non-malignant is used here to encompass benign neoplasms and non-neoplastic mass lesions.

Materials and Methods

Records of the Queen Mother Hospital for Animals of The Royal Veterinary College in the period 2008-2014 were reviewed in a retrospective, cross-sectional study. Inclusion criteria were dogs that had a dual-phase, post-contrast CT study of the abdomen and subsequent histopathologic diagnosis of an hepatic or splenic mass. For the purposes of this study, mass was defined as a focal parenchymal lesion with displacement of adjacent structures and >10mm maximal transverse dimension. Dogs in which the CT examination and histological diagnosis were separated by 30 days or more were not included. Dogs in which diagnosis was based on cytology were not included.

CT scans were routinely performed in anaesthetized or sedated patients in sternal recumbency. All scans were obtained using a 16-slice MDCT scanner (MX 8000 IDT, Philips Medical Systems, Cleveland, USA). CT settings were helical acquisition, slice thickness 3mm, image reconstruction interval 1.5mm, helical pitch 0.688, tube rotation time 0.75s, x-ray tube current 150 mAs, x-ray tube potential 120kVp, field of view 400mm, matrix 512x512, and medium frequency ('soft tissue') reconstruction algorithm. Scans were performed in a cranial to caudal direction starting at the cardiac apex and terminating at the mid-femoral diaphysis.

Post-contrast CT scanning followed a dual-phase protocol introduced at our institution in 2008 for all dogs having abdominal CT examinations, except those with suspected portosystemic shunting. In each dog, 2ml/kg of iohexol 300mg/ml (Omnipaque 300, GE Healthcare, Oslo, Norway) was injected intravenously at 2ml/s (maximum pressure 150 psi) using a power injector (Stellant, Medrad Inc., Pennsylvania, USA) via a catheter in a cephalic or saphenous vein. Images of the whole abdomen were acquired prior to, within 30 seconds (early phase) and at least 60 seconds (delayed phase) following the start of intravenous injection of contrast, as described for dual-phase CT protocols for humans.[8,14-17] . The variable need for entry of the anesthetist into the CT room to examine their patient after the first post-contrast CT acquisition contributed to variations in the timing of the second post-contrast acquisition, which was initiated only after all personnel had exited the CT room.

CT images were reviewed by a Board-certified radiologist (PM) who was blinded to the final diagnosis. The reviewer evaluated only transverse images. Where multiple CT examinations were available for a dog, only the first examination was reviewed because later studies were generally obtained after biopsy of the mass, surgery or other treatment. Dogs were excluded if review of their CT images found multiple hepatic or splenic lesions with differing imaging

features. This was considered necessary in order to be able to relate the biopsy result in each dog with the corresponding features in CT images.

The following patient information was recorded: Site of mass (hepatic/splenic), diagnosis, age at time of study, breed, sex and weight. The catheter site used for contrast administration was also recorded.

Commercially available software (OsiriX, Pixmeo, SARL, 266, Rue de Bernex, CH-1233, Bernex, Switzerland) was used for image review and analysis. Window width was 400 Hounsfield units (HU), window level was 60 HU. Aortic and portal vein attenuation in early and delayed phase images was recorded. The maximal transverse dimension of each mass was measured using electronic calipers. Normal attenuation value of hepatic/splenic parenchyma was determined based on the mean of three 20mm^2 regions of interest (ROIs). Attenuation of masses was measured using the maximum circular ROI that could be fitted to each mass. Identical ROIs were used for measurements in pre-contrast images and early and delayed phase images. In pre-contrast CT images, hyperattenuation was defined as at least 10HU greater than normal parenchyma; hypoattenuation was defined as at least 10HU less than normal parenchyma; isoattenuation was defined as attenuation within 10 HU of normal parenchyma.[22]

The following quantitative variables were recorded: maximal transverse dimension of the mass (mm), mean hepatic/splenic parenchymal attenuation pre-contrast (HU), attenuation of lesion pre-contrast (HU), relative attenuation of lesion (lesion attenuation – mean hepatic/splenic parenchymal attenuation), attenuation of lesion in early phase (HU), early enhancement of lesion (attenuation of lesion in early phase – attenuation of lesion pre-contrast), delayed attenuation of lesion (HU), delayed enhancement of lesion (delayed attenuation of lesion – pre-contrast attenuation of lesion) and contrast accumulation within the lesion (delayed enhancement of

lesion – early enhancement of lesion). Slight enhancement was defined as <10HU; marked enhancement was defined as ≥ 10 HU.

The following subjective categorical variables were recorded: appearance of the margin of mass (distinct/indistinct), parenchymal homogeneity of mass in pre-contrast images (homogenous/heterogeneous), hepatic or splenic capsular distortion as a result of mass effect (yes/no), pattern of contrast enhancement (generalized /peripheral), presence of peritoneal fluid (yes/no), mineralization within mass (yes/no), and presence of abnormal hepatic and/or splenic lymph nodes (yes/no). [22,24]

Tissue for histological analysis was acquired by ultrasound-guided tissue core biopsy using 14G or 18G needles, wedge resection at exploratory laparotomy, partial hepatectomy or splenectomy. All histopathologic specimens were reviewed by a Board-certified pathologist (SLP).

Normality of continuous data was assessed by visual inspection and the Kolmogorov-Smirnov test. Data did not conform to a Normal distribution, hence differences were tested using a Mann-Whitney U-test. Likelihood ratios (LR) and their 95% confidence intervals (95% CI) were calculated to describe the strength of associations between categorical features and malignancy. Statistical tests were performed using commercially available (SPSS version 16, SPSS Inc., Chicago, Illinois, USA) and online software (Stats calculator, Centre for Evidence Based Medicine, Toronto, Canada, http://ktclearinghouse.ca/cebm/practise/ca/calculators/statscalc).

Results

Fifty-two dogs with 55 masses were included: 24 had an hepatic mass, 31 had a splenic mass, and 3 dogs had both a hepatic and splenic mass (2 dogs had primary splenic hemangiosarcoma

with metastasis to the liver, and one dog had primary splenic spindle cell sarcoma with metastasis to the liver). Dogs were 11 mixed breed, 7 Labrador retrievers, 5 German shepherd dogs, 3 Flat-coated retrievers, 2 Boxers, 2 Golden retrievers, 2 Schnauzers and 2 West highland white terriers and 1 each of the following breeds: Beagle, Bernese mountain dog, Border collie, Border terrier, Bulldog, Cairn terrier, Chow Chow, Cocker spaniel, Dachshund, Dog-de-Bordeaux, English springer spaniel, Irish setter, Lhasa Apso, Lurcher, Papillon, Rhodesian ridgeback, Tibetan terrier, and Yorkshire terrier. Thirty-two dogs were male (20 neutered) and 20 were female (19 neutered). Their median age was 10 years (range 3 – 15 years).

Median patient weight was 24kg (4-54kg). Cephalic catheters were used to administer contrast in 49 (94%) dogs and saphenous catheters were used in 3 (6%) dogs. In every dog, acquisition of early phase images occurred less than 30s after the administration of contrast. The median delay between contrast administration and delayed phase image acquisition was 120s (range 62-465s). Median aortic attenuation was 207HU (range 110-607HU) in early phase images and 130HU (range 49-219HU) in delayed phase images. Median portal attenuation was 155HU (range 48-315HU) in early phase images and 129HU (range 49-226HU) in delayed phase images.

Of the 24 hepatic masses, 14 (58%) were malignant (9 carcinoma, 3 metastatic splenic hemangiosarcomas, 1 metastatic splenic spindle cell sarcoma, 1 hemangiosarcoma) and 10 (42%) were non-malignant (6 adenoma, 3 nodular hyperplasia, 1 bile duct hyperplasia). Of the 31 splenic masses, 18 (58%) were malignant (13 hemangiosarcomas, 3 sarcomas, 1 histiocytic sarcoma, 1 metastatic renal nephroblastoma) and 13 (42%) were non-malignant (8 hematomas, 5 nodular hyperplasia).

There were no significant differences in quantitative or categorical CT image data between malignant and non-malignant masses in the liver (Tables 1 & 2) or spleen (Tables 3 & 4).

Similarly, there was substantial overlap in the pre- and post-contrast imaging features of the most prevalent types of hepatic and splenic masses (Tables 5 & 6, respectively). Recognizing that the wide variation in vascular attenuation observed in this study could potentially obscure differences between types of hepatic and splenic masses, the analyses were repeated after removing data for 8 dogs with delayed phase images >240s after administration of contrast and the 3 dogs with saphenous catheters; however, the significance of results of statistical tests were unchanged, hence these dogs were retained.

Hepatic carcinoma, hepatic nodular hyperplasia and hepatic adenoma most frequently had lower attenuation and were heterogeneous compared to surrounding hepatic parenchyma in pre-contrast images, and the majority of these lesions showed marked, generalized enhancement in the early phase images that persisted in the delayed phase (Figure 1). The majority of hepatic masses lacked contrast accumulation. Splenic hemangiosarcoma, hematoma, and nodular hyperplastic masses were mostly slightly heterogeneous with mean attenuation similar to adjacent splenic parenchyma in pre-contrast images. In early and delayed phase images, splenic hemangiosarcoma and nodular hyperplastic lesions most frequently showed marked, diffuse enhancement in early phase images that persisted in delayed images. Most splenic hematomas had slight enhancement in early phase images but marked enhancement in delayed images, as a result of contrast accumulation (Figure 2).

Discussion

In the present study, no CT features were found to be significantly associated with malignant or non-malignant hepatic masses. The most prevalent of the hepatic masses studied – hepatic carcinoma, adenoma, and nodular hyperplasia – each had a similar range of CT features. Most

frequently these masses were heterogeneous compared to surrounding hepatic parenchyma in pre-contrast images, and the majority of these lesions showed marked, generalized enhancement in the early phase images that persisted in the delayed phase.

The present study employed a dual-phase CT protocol comparable to dual-phase CT protocols used for assessment of hepatic lesions in humans.[8,14-17] There are no other reports of veterinary studies that used the same dual-phase CT protocol for assessment of hepatic or splenic masses. Two recent studies employed triple-phase CT protocols with images acquired at 13-20s (HAP), 30-40s (early PVP) and 120s (delayed phase) following contrast administration.[21,22] Compared to these, the early phase in the dual-phase CT protocol is similar to the early PVP, and the delayed phase is similar to the delayed phase of triple-phase CT. Omission of HAP images is unlikely to significantly reduce ability to distinguish types of hepatic mass because in previous series a large proportion of masses had similar degrees of post-contrast enhancement in HAP and PVP images.[21,22]

In HAP images, enhancement of tissues represents contrast medium within the intravascular space and the degree of enhancement is determined primarily by the hepatic arterial blood flow.[10] Subsequent tissue enhancement represents additional delivery of contrast via the portal vessels and passage of contrast material from the intravascular to the extravascular space across the capillary basement membrane, hence the degree of tissue enhancement also depends on blood volume and permeability of capillaries.[10] Based on previous studies in humans[16] and dogs[21,22], hepatic masses containing relatively well-differentiated hepatocytes tend to enhance most strongly in HAP images, probably reflecting both their relatively abundant arterial blood flow and lack of necrotic or hemorrhagic (vascular) components. However, these observations do not represent differences that enable different histologic types of hepatic masses to be distinguished

clinically because – as observed in the present study – marked enhancement in early phase images occurs with malignant neoplasia, such as hepatocellular carcinoma, and with non-malignant lesions, such as hepatic adenoma and nodular hyperplasia.[21,22]

Differentiation of hepatocellular carcinoma from hepatic adenoma and hepatic nodular hyperplasia depends on histopathology.[25,26] Hepatic carcinomas contain pleomorphic hepatocytes arranged in irregular trabeculae separated by loose vascular (sinusoidal) spaces and a variable amount of fibrous connective tissue, and exhibit varying degrees of internal hemorrhage and necrosis. Sinusoidal invasion beyond a fibrous capsule or pseudocapsule and/or intrahepatic metastasis are the key histologic criteria of hepatocellular carcinoma, but the former may be subtle and/or focal in a well-differentiated tumor. Hepatic adenomas contain well-differentiated hepatocytes in rough cords and near normal architecture, but lack portal triads or central veins. Hepatic hyperplastic nodules contain well-differentiated hepatocytes with slightly reduced numbers of portal triads or central veins compared to normal liver, but with a distorted lobular architecture and frequent vacuolar changes representing glycogen or lipid accumulation. It is important to note that the histologic diagnosis of these hepatic lesions depends on cellular and architectural features that exist in a spectrum of severity in which the boundaries between well-differentiated hepatocellular carcinoma and adenoma, and between adenoma and nodular hyperplasia, are not always clearly defined. [25,26] Consequently, there will be a limit to the degree that CT features, which primarily represent non-specific gross pathologic features, such as pseudoencapsulation, hemorrhage and necrosis[27], can be used to deduce the histologic diagnosis of hepatic masses. The lack of significant differences in CT features associated with malignant or non-malignant hepatic masses observed in the present study likely reflects this limitation.

Splenic hemangiosarcoma, hematoma, and nodular hyperplastic masses also exhibited variable CT features. In pre-contrast images, most splenic masses were slightly heterogeneous, with median attenuation similar to adjacent parenchyma. In early and delayed phase images, splenic hemangiosarcoma and nodular hyperplastic lesions most frequently showed marked, generalized enhancement in early phase images that persisted in delayed phase images. These results do not corroborate those of a previous study in which malignancy of a splenic mass was associated with hypoattenuation in pre-contrast images and minimal contrast accumulation in post-contrast images, with 55HU in post-contrast images representing a threshold value to distinguish malignant (<55HU) from non-malignant (>55HU) masses.[23] In the present study, there was no difference in median enhancement in delayed phase images between malignant and non-malignant splenic masses with both types of mass having a wide range of post-contrast attenuation values that spanned 55HU.

Most splenic hematomas had slight enhancement in early phase images, and all hematomas had contrast accumulation in delayed phase images. This feature could represent active hemorrhage occurring during the CT examination[20], but was not unique to hematomas as it was also observed in the majority of hemangiosarcomas. Marked focal contrast accumulation in hemangiosarcomas was reported recently, possibly representing contrast medium in wide vascular spaces within these tumors.[28] Splenic malignant neoplasms, most of which are hemangiosarcomas, frequently contain extensive hemorrhage or necrosis and so may be difficult to distinguish from hematoma on the basis of either gross or histologic examination.[3,29-31] As noted above with respect to hepatic masses, other imaging techniques that depict vascularity of tissues also have limited potential to distinguish the histologic types of splenic mass.[32-35]

The present study is subject to patient selection bias because of the requirement for histologic diagnosis, which necessitated biopsy. Lesions suspected to be malignant were more likely to be considered priorities for biopsy than lesions suspected to be benign (e.g. because they appeared unrelated to clinical signs or were known to be static), hence benign masses may be underrepresented compared to the overall number of masses encountered in the clinical setting. Similarly, hepatic or splenic masses that are routinely diagnosed in our hospital on the basis of cytologic and flow cytometric findings, primarily round cell neoplasms, were not included.

Patient factors, the method of contrast injection, and timing of CT image acquisition affect the magnitude of enhancement of tissues observed following intravenous contrast administration.[11,17] Patient body weight and cardiac output will affect the time of the peak plasma concentration of contrast medium even if the iodide concentration of the contrast agent, its dose and rate of injection all remain constant.[11,17] Injection duration is the most important injection factor to be considered for determining CT scan timing because it directly affects the time to peak contrast enhancement in an organ or vessel.[11,17] With an increased duration of contrast injection, the time to peak opacification increases. For example, the protocol used in the present study with an injection rate 2ml/s will result in a slightly later peak hepatic arterial opacification than that observed using 5ml/s.[36] Also, in the present study, both early and delayed post-contrast images were acquired in a cranial to caudal direction whereas the early phase may also be acquired in a caudal to cranial direction.[36]

Variations in the degree of vascular enhancement were evident in the present study. Although acquisition of early phase images always occurred less than 30s after the start of intravenous injection of contrast, there was a wide range of values for aortic attenuation in early phase images. This range will include animals imaged near the peak of aortic enhancement and those

306 imaged after the peak, when portal vein attenuation is rising. Early phase images to examine the
307 liver are acquired after peak arterial enhancement because contrast accumulation representing the
308 hepatic arterial supply to parenchymal lesions will necessarily continue after peak arterial
309 attenuation.[8,11,14] This is a different technique to that employed for studies focused on vascular
310 anatomy, such as investigation of portosystemic shunting[37,38], when it is important to time the CT
311 acquisition to coincide with peak vascular attenuation. The finding that portal attenuation was
312 increased in early phase images in the present study is to be expected because portal attenuation
313 normally exceeds hepatic arterial attenuation within 30s in anaesthetized dogs.[36] Conversely, the
314 finding of persistently high aortic attenuation in some delayed phase images suggests that other
315 factors, such as slow circulation time and/or reduced renal excretion of contrast, possibly
316 associated with anesthesia, may have affected some dogs. The rate of change of vascular and
317 parenchymal contrast enhancement is most rapid soon after contrast injection and slows
318 subsequently[11], hence variations in timing of delayed phase images may have less effect on
319 contrast accumulation in tissues than variations during the early phase. In the present study,
320 repeating the analyses after removing data for dogs with delayed phase images >240s after
321 administration of contrast did not alter the significance of results of statistical tests. Nevertheless,
322 patient factors and variations in timing of CT image acquisition will have affected the degree of
323 enhancement of masses in post-contrast images, and may have contributed to the overlap
324 observed between CT features of the hepatic and splenic masses in the present study.

325 Automatic triggering of the CT acquisition, based on attenuation within a region of interest (e.g.
326 the abdominal aorta) exceeding a threshold, is a software feature of CT scanners that can be used
327 to increase precision in the timing of post-contrast images. Automatic triggering of the CT
328 acquisition was not utilized for the present study. A test bolus technique has been employed for

dual-phase CT studies of dogs with suspected portosystemic shunting[37,38] and insulinoma.[39] This technique enables optimal timing of the vascular phases of a multi-phase CT acquisition, but it requires an additional scan and image data analysis[36], so is more time-consuming, which is disadvantageous for routine abdominal examinations. Recently, dual-phase CT of conscious sedated and unsedated dogs was described using fixed time delays equal to injection duration for HAP images and 40s for PVP images.[40] Quality of images obtained was judged to be good or excellent in 72% dogs. Poor quality scans were primarily the result of motion blur or other artifacts; in only one instance was the HAP missed.[40] Use of fixed time delays is a pragmatic approach for routine abdominal CT studies, although test bolus or automatic triggering methods enable CT scans to be acquired with a more accurately defined vascular phases.

On the basis of these results, it may be concluded that the dual-phase protocol we employed for CT of dogs with hepatic or splenic masses provides limited specific diagnostic information. Variability in contrast delivery to masses and relatively low numbers of subjects limit the statistical power of the study. It is possible that significant associations between CT signs and diagnosis would be detected if larger numbers of subjects were studied under more consistent conditions, hence further studies are warranted.

345 Table 1. Size, attenuation, and post-contrast enhancement characteristics of malignant and non-

346 malignant hepatic masses

	Malignant (n=14)	Non-malignant (n=10)	P-value
Maximum transverse dimension (mm)	61 (15 - 695)	60 (32 - 345)	0.95
Mean hepatic parenchymal attenuation (HU)	60 (53 - 75)	71 (49 - 79)	0.13
Pre-contrast attenuation (HU)	46 (32 - 68)	60 (26 - 69)	0.32
Relative attenuation (HU)	-13 (-28 - -3)	-16 (-35 - -2)	0.75
Early post-contrast attenuation (HU)	83 (42 - 152)	95 (22 - 125)	0.77
Early enhancement (HU)	31 (2 - 113)	42 (-4 - 65)	0.77
Delayed post-contrast attenuation (HU)	77 (42 - 120)	99 (25 - 121)	0.62
Delayed enhancement (HU)	27 (1 - 81)	34 (-1 - 53)	0.98
Contrast accumulation (HU)	-3 (-32 - 20)	-5 (-23 - 52)	0.64

347

348 Values are median (range); HU, Hounsfield units

349 Table 2. Descriptive features of malignant and non-malignant hepatic masses

	Malignant (n=14)	Non-Malignant (n=10)	LR	95% CI
Distinct margins	7 (64%)	4 (36%)	2.0	0.5-3.1
Indistinct margins	7 (54%)	6 (46%)	0.8	0.4-1.7
Homogeneous	8 (67%)	4 (33%)	1.4	0.6-3.5
Heterogeneous	6 (50%)	6 (50%)	0.7	0.3-1.6
Capsular distortion	12 (60%)	8 (40%)	1.1	0.7-1.6
Generalized enhancement	12 (60%)	8 (40%)	1.1	0.7-1.6
Peripheral enhancement	2 (50%)	2 (50%)	0.7	0.1-4.3
Peritoneal fluid	4 (57%)	3 (43%)	0.9	0.3-3.2
Mineralization within mass	3 (75%)	1 (25%)	2.1	0.3-17.7
Abnormal local lymph nodes	11 (65%)	6 (35%)	1.3	0.7-2.3

350

351 LR, Likelihood ratio associated with malignancy

352

353 Table 3. Size, attenuation, and post-contrast enhancement characteristics of malignant and

354 non-malignant splenic masses

	Malignant (n=18)	Non-malignant (n=13)	P-value
Maximum transverse dimension (mm)	69 (12 - 204)	73 (19 - 173)	0.28
Mean splenic parenchymal attenuation (HU)	53 (19 - 76)	61 (35 - 67)	0.18
Pre-contrast attenuation (HU)	43 (-3 - 55)	49 (12 - 73)	0.12
Relative attenuation (HU)	-6 (-30 - 35)	-2 (-52 - 20)	0.47
Early post-contrast attenuation (HU)	63 (0 - 106)	73 (14 - 119)	0.36
Early enhancement (HU)	20 (0 - 69)	20 (-3 - 64)	0.67
Delayed post-contrast attenuation (HU)	73 (0 - 120)	81 (16 - 112)	0.98
Delayed enhancement (HU)	28 (0 - 82)	30 (4 - 57)	0.50
Contrast accumulation (HU)	10 (-11 - 56)	3 (-7 - 27)	0.56

355

356 Values are median (range); HU, Hounsfield units

357

358 Table 4. Descriptive features of malignant and non-malignant splenic masses

	Malignant (n=18)	Non-Malignant (n=13)	LR	95% CI
Distinct margins	6 (43%)	8 (57)	0.5	0.2-1.2
Indistinct margins	12 (71%)	5 (29%)	1.7	0.8-3.7
Homogeneous	5 (56%)	4 (44%)	0.9	0.3-2.7
Heterogeneous	13 (59%)	9 (41%)	1.0	0.6-1.7
Capsular distortion	17 (59%)	12 (41%)	1.0	0.8-1.2
Generalized enhancement	16 (57%)	12 (43%)	0.9	0.8-1.2
Peripheral enhancement	2 (67%)	1 (33%)	1.4	0.1-14.3
Peritoneal fluid	12 (60%)	8 (40%	1.1	0.6-1.9
Mineralization within mass	1 (100%)	0 (0%)	2.2	0.1-50.3
Abnormal local lymph nodes	6 (55%)	5 (45%)	0.9	0.3-2.2

359

360 LR, Likelihood ratio associated with malignancy

361

362 Table 5. Pre- and post-contrast features of hepatocellular carcinoma, adenoma, and nodular

363 hyperplasia

	Hepatocellular carcinoma (n=9)	Adenoma (n=6)	Nodular hyperplasia (n=3)
Pre Contrast			
Hypoattenuation	5 (56%)	5 (83%)	2 (67%)
Isoattenuation	4 (44%)	1 (17%)	1 (33%)
Homogenous	3 (33%)	2 (33%)	2 (67%)
Heterogeneous	6 (66%)	4 (66%)	1 (33%)
Early Phase			
Slight enhancement	2 (22%)	1 (17%)	2 (67%)
Marked enhancement	7 (78%)	5 (83%	1 (33%)
Generalized	7 (78%)	6 (100%)	2 (67%)
Peripheral	2 (22%)	0	1 (33%)
Late Phase			
Slight enhancement	2 (22%)	0	2 (67%)
Marked enhancement	7 (78%)	6 (100%)	1 (33%)
Contrast accumulation	2 (22%)	0	1 (33%)
Lack of contrast accumulation	7 (78%)	6 (100%)	2 (67%)

364

365 Table 6. Pre- and post-contrast features of splenic hemangiosarcoma, hematoma, and nodular

366 hyperplasia

	Hemangiosarcoma (n=13)	Hematoma (n=8)	Nodular hyperplasia (n=5)
Pre Contrast			
Hypoattenuation	4 (31%)	2 (25%)	3 (60%)
Isoattenuation	8 (62%)	5 (63%)	2 (40%)
Hyperattenuation	1 (7%)	1 (12%)	0
Homogenous	4 (31%)	3 (38%)	1 (20%)
Heterogeneous	9 (69%)	5 (62%)	4 (80%)
Early Phase			
Slight enhancement	2 (15%)	5 (62%)	0
Marked enhancement	11 (85%)	3 (38%)	5 (100%)
Generalized	12 (92%)	7 (88%)	5 (100%
Peripheral	1 (7%)	1 (12%)	0
Late Phase			
Slight enhancement	1 (7%)	3 (38%)	0
Marked enhancement	12 (92%)	5 (62%)	5 (100%
Contrast accumulation	10 (77%)	8 (100%)	2 (40%)
Lack of contrast accumulation	3 (23%)	0	3 (60%)

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References

1. Patnaik AK, Hurvitz AI, Lieberman PH, Johnson GF. Canine hepatocellular carcinoma. Vet Pathol 1981; 18: 427-438.

2. Balkman C. Hepatobiliary neoplasia in dogs and cats. Vet Clin North Am: Small Anim Pract 2009; 39: 617–625.

3. Prymak C, McKee LJ, Goldschmidt MH, Glickman LT. Epidemiologic, clinical, pathologic, and prognostic characteristics of splenic hemangiosarcoma and splenic hematoma in dogs: 217 cases (1985). J Am Vet Med Assoc 1988; 193:706-712.

4. Spangler WL, Culbertson MR. Prevalence, type, and importance of splenic diseases in dogs: 1,480 cases (1985-1989). J Am Vet Med Assoc 1992; 200: 829-834.

5. Bergman JR. Nodular hyperplasia in the liver of the dog: an association with changes in the Ito cell population. Vet Pathol 1985; 22: 427-438.

6. Stowater JL, Lamb CR, Schelling SH. Ultrasonographic features of canine hepatic nodular hyperplasia. Vet Radiol 1990;31:268-272.

7. Johnson KA, Powers BE, Withrow SJ, Sheetz MJ, Curtis CR, Wrigley RH. Splenomegaly in dogs. Predictors of neoplasia and survival after splenectomy. J Vet Internal Med 1989; 3:160-166.

8. Sahani DV, Singh AH. Dual-phase liver MDCT. In: Kalra MK, Saini S, Rubin GD (Eds). MDCT From Protocol to Practice. Milan: Springer, 2008, pp83-92.

9. Ji H, McTavish JD, Mortele KJ, Wiesner W, Ros PR. Hepatic imaging with multidetector CT. Radiographics 2001;21:S71-S80.

10. Miles, KA. Tumour angiogenesis and its relation to contrast enhancement on computed tomography: a review. Eur J Radiol 1999; 30:198-205.

11. Bae KT. Principles of contrast medium delivery and scan timing in MDCT. In: Kalra MK, Saini S, Rubin GD (Eds). MDCT From Protocol to Practice. Milan: Springer, 2008, pp10-24.

12. Hwang GJ, Kim MJ, Yoo HS, Lee JT. Nodular hepatocellular carcinomas: Detection with arterial-, portal-, and delayed-phase images at spiral CT. Radiology 1997; 202:383-388.

13. Murakami T, Kim T, Takamura M, et al. Hypervascular hepatocellular carcinoma: detection with double arterial phase multidetector helical CT. Radiology 2001; 218:763-767.

14. Oliver JH, Baron RL, Federle MP, Jones BC, Sheng R. Hypervascular liver metastases: do unenhanced and hepatic arterial phase CT images affect tumor detection? Radiology 1997; 205:709-715.

15. Ichikawa T, Kitamura T, Nakajima H, Sou H, Tsukamoto T, Ikenaga S, et al. Hypervascular hepatocellular carcinoma: can double arterial phase imaging with multidetector CT improve tumor depiction in the cirrhotic liver? Am J Roentgenol 2002;179:751-758.

16. Baron RL, Oliver JH, Dodd GD, Nalesnik M, Holbert BL, Carr B. Hepatocellular carcinoma: evaluation with biphasic, contrast-enhanced, helical CT. Radiology 1996; 199:505-511.

17. Bae KT. Intravenous contrast medium administration and scan timing at CT: considerations and approaches. Radiology 2010;256:32-61.

18. Ichikawa T, Erturkb SM, Arakia T. Multiphasic contrast-enhanced multidetector-row CT of liver: Contrast-enhancement theory and practical scan protocol with a combination of fixed injection duration and patients' body-weight-tailored dose of

contrast material. Eur J Radiol 2006;58: 165–176.

19. Chan MG, Cassidy FH, Andre MP, Chu P, Aganovic L. Delayed imaging in routine CT examinations of the abdomen and pelvis: is it worth the additional cost of radiation and time? Am J Roentgenol 2014;202:329-335.

20. Vasanawala SS, Desser T. Value of delayed imaging in MDCT of the abdomen and pelvis. Am J Roentgenol 2006;187:154-163

21. Fukushima K, Hideyuki K, Ohno K, Takahashi M, Nakashima K, Fujino Y, et al. CT Characteristics of primary hepatic mass lesions in dogs. Vet Radiol Ultrasound 2012; 53: 252–257.

22. Kutara K, Seki M, Ishikawa C, Sakai M, Kagawa Y, Iida G, et al. Triple-phase helical computed tomography in dogs with hepatic masses. Vet Radiol Ultrasound 2014; 55:7–15.

23. Fife WD, Samii VF, Drost T, Mattoon JS, Hoshaw-Woodard S. Comparison between malignant and nonmalignant splenic masses in dogs using contrast enhanced computed tomography. Vet Radiol Ultrasound 2004; 45: 289–297.

24. Beukers M, Vilaplana Grosso F, Voorhout G. Computed tomographic characteristics of presumed normal canine abdominal lymph nodes. Vet Radiol Ultrasound 2013; 54: 610–617.

25. Cullen JM, Brown DL. Hepatobiliary system and exocrine pancreas. In: Zachery JF, McGavin MD (Eds). Pathologic Basis of Veterinary Disease, 5th edition. St. Louis: Elsevier, 2012, pp442-445.

26. Stalker MJ, Hayes MA. Liver and biliary system. In: Maxie MG (Ed). Jubb, Kennedy, and Palmer's Pathology of Domestic Animals, 5th edition, vol.2. St. Louis: Elsevier, 2007, pp382-388.

27. Gregori T, Mantis P, Benigni L, Priestnall S, Lamb CR. Comparison of computed tomographic and pathologic findings in 17 dogs with primary adrenal neoplasia. Vet Radiol Ultrasound 2015;56:153-159.

28. Fukuda S, Kobayashi T, Robertson ID, Oshima F, Fukazawa E, Nakano Y, et al. Computed tomographic features of canine nonparenchymal haemangiosarcoma. Vet Radiol Ultrasound 2014; 55: 374–379.

29. Day MJ, Lucke VM, Pearson H. A review of pathological diagnoses made from 87 canine splenic biopsies. J Small Anim Pract 1995; 36: 426-433.

30. Maxie MG, Robinson WF. Cardiovascular system. In: Maxie MG (Ed). Jubb, Kennedy, and Palmer's Pathology of Domestic Animals, 5th edition, vol.3. St. Louis: Elsevier, 2007, pp102-105.

31. Mallinckrodt MJ, Gottfried SD. Mass-to-splenic volume ratio and splenic weight as a percentage of body weight in dogs with malignant and benign splenic masses: 65 cases (2007-2008). J Am Vet Med Assoc 2011;239:1325-1327.

32. Taeymans O, Penninck DG. Contrast enhanced sonographic assessment of feeding vessels as a discriminator between malignant vs. benign focal splenic lesions. Vet Radiol Ultrasound 2011; 52:457–461.

33. Rossi F, Leone VF, Vignoli M, Laddaga E, Terragni R. Use of contrast-enhanced ultrasound for characterization of focal splenic lesions. Vet Radiol Ultrasound 2008;49:154-164.

34. Ivancic M, Long F, Seiler GS. Contrast harmonic ultrasonography of splenic masses and associated liver nodules in dogs. J Am Vet Med Assoc 2009;234: 88-94.

35. Nakamura K, Sasaki N, Murakami M, Kumara WRB, Ohta H, Yamasaki M, et al. Contrast-enhanced ultrasonography for characterization of focal splenic lesions in dogs.

463 J Vet Internal Med 2010;24:1290-1297.

464 36. Zwingenberger AL, Schwartz T. Dual-phase CT angiography of the normal canine
465 portal and hepatic vasculature. Vet Radiol Ultrasound 2004; 45: 117-124.

466 37. Zwingenberger AL, Schwarz T, Saunders HM. Helical computed tomographic
467 angiography of canine portosystemic shunts. Vet Radiol Ultrasound 2005;46: 27-32.

468 38. Park K, Yeon S, Lee H. Hepatic arteriovenous fistula in four dogs: usefulness of dual-
469 phase computed tomographic angiography. J Biomed Res 2012;13:195-199.

470 39. Mai W, Cáceres AV. Dual-phase computed tomographic angiography in three dogs
471 with pancreatic insulinoma. Vet Radiol Ultrasound 2008; 49: 141–148.

472 40. Shanaman MM, Hartman SK, O'Brien RT. Feasibility for using dual-phase contrast-
473 enhanced multi-detector helical computed tomography to evaluate awake and sedated
474 dogs with acute abdominal signs. Vet Radiol Ultrasound 2012;53: 605–612.

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Legends

Figure 1. Examples of dual-phase CT images of hepatic masses in dogs. A) Hepatocellular carcinoma; B) adenoma (between arrows); C) nodular hyperplasia (between arrowheads). In each instance images are pre-contrast (left), early phase (middle) and delayed phase (right). Each of these lesions has slightly lower attenuation and is heterogeneous compared to surrounding hepatic parenchyma in pre-contrast images, and show marked, generalized enhancement in the early phase images that persists in delayed phase images. Multiple masses with similar features are present throughout the liver in the dog with hepatocellular carcinoma. Window 350HU, level 50HU.

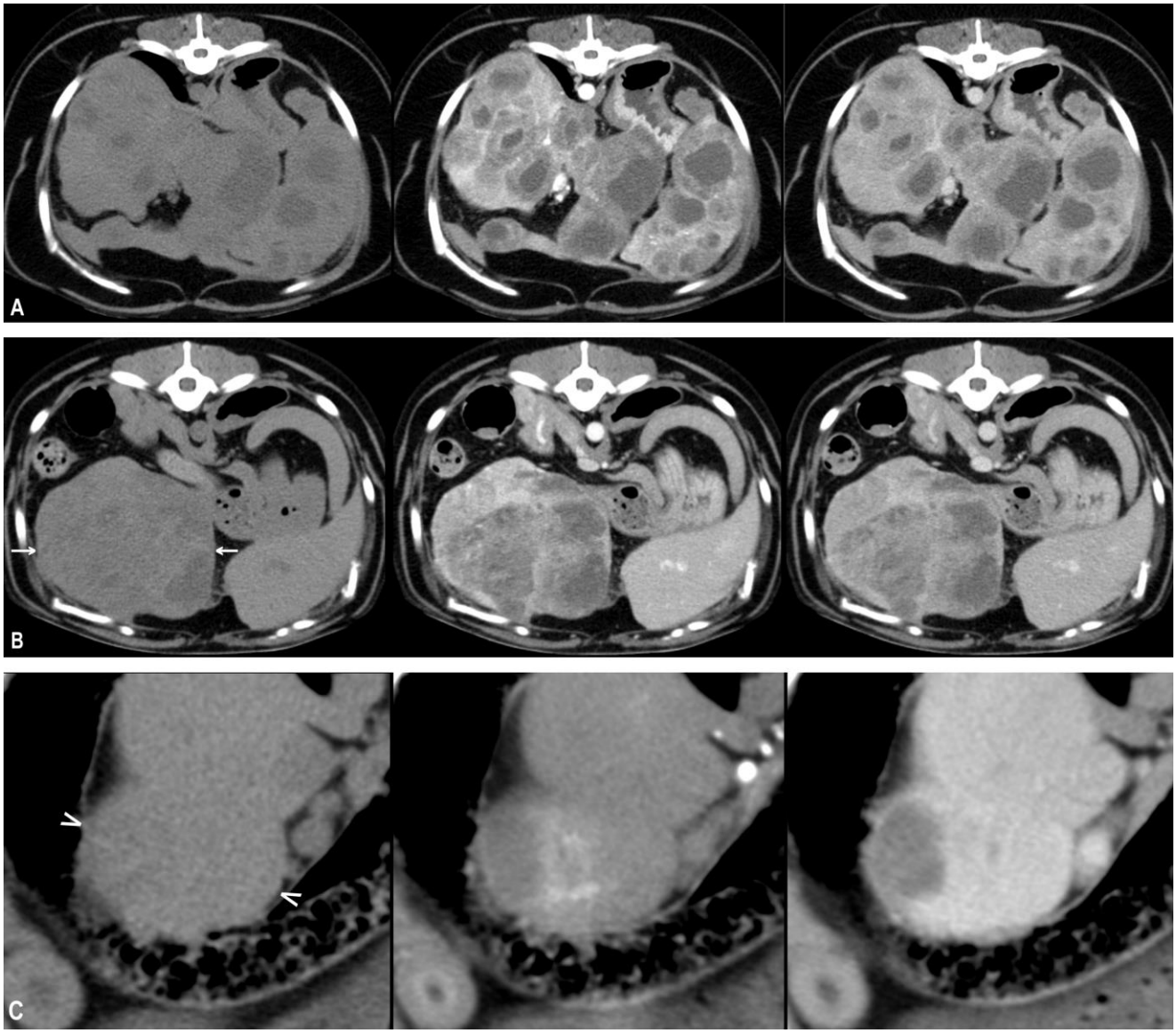

Figure 2. Examples of multi-phase CT images of splenic masses in dogs. A) hemangiosarcoma (between arrowheads); B) hematoma (between arrows); C) nodular hyperplasia (within dotted line). Each of these lesions has slightly lower attenuation and is heterogeneous compared to surrounding hepatic parenchyma in pre-contrast images, and show marked, generalized heterogeneous enhancement in early phase images that increases in delayed phase images as a result of contrast accumulation. Foci of marked local contrast accumulation are evident in the hyperplastic mass (arrowhead) compatible with contrast in large vascular spaces.[28] Multiple segmental lesions affecting the kidneys in the dog with hemangiosarcoma are compatible with infarcts. H, peritoneal hematoma. Window 350HU, level 50HU.